Cell, Volume *179*

Supplemental Information

Rare Pulmonary Neuroendocrine Cells

Are Stem Cells Regulated by Rb, p53, and Notch

Youcef Ouadah, Enrique R. Rojas, Daniel P. Riordan, Sarah Capostagno, Christin S. Kuo, and Mark A. Krasnow

Table S1 (related to Figs. 1, S1). Proliferative NE cells in Rainbow-labeled NEBs one week after airway injury[1]

NEB I.D.	Injury[2]	No. of CGRP$^+$ cells	mCerulean$^+$ cells		mOrange$^+$ cells		mCherry$^+$ cells		EGFP$^+$ cells		Inferred min. NE$^{stem\ 3}$	Inferred max. NE$^{stem\ 4}$
			EdU$^+$	Total	EdU$^+$	Total	EdU$^+$	Total	EdU$^+$	Total		
NEB1	–	17	0	9	0	3	0	2	0	3	0	0
NEB2	–	20	0	8	0	1	0	2	0	10	0	0
NEB3	–	22	0	9	0	3	0	5	0	5	0	0
NEB4	–	23	0	12	0	3	0	3	0	5	0	0
NEB5	–	22	0	11	0	2	0	0	0	9	0	0
NEB6	–	16	0	8	0	3	0	3	0	2	0	0
NEB7	–	20	0	14	0	1	0	3	0	2	0	0
NEB8	–	27	0	15	0	5	0	1	0	6	0	0
NEB9	–	23	0	12	0	3	0	2	0	6	0	0
NEB10	–	37	0	18	0	10	0	4	0	5	0	0
NEB11	–	24	0	17	0	3	0	1	0	3	0	0
NEB12	–	43	0	25	0	9	0	5	0	4	0	0
NEB13	–	13	0	7	0	4	0	0	0	2	0	0
NEB14	–	28	0	14	0	8	0	2	0	4	0	0
NEB15	–	40	0	23	0	4	0	8	0	5	0	0
NEB16	–	10	0	5	0	3	0	0	0	2	0	0
NEB17	–	17	0	7	0	5	0	2	0	3	0	0
NEB18	–	20	0	15	0	1	0	2	0	2	0	0
NEB19	–	24	0	11	0	6	0	3	0	4	0	0
NEB20	–	36	0	21	0	3	0	5	0	7	0	0
NEB21	–	25	0	16	0	3	0	2	0	4	0	0
NEB22	–	14	0	8	0	4	0	2	0	0	0	0
NEB23	–	26	0	18	0	4	0	2	0	2	0	0
NEB24	+	45	2	21	0	3	3	0	4	18	2	3
NEB25	+	24	2	14	0	2	1	0	0	7	1	1
NEB26	+	22	2	11	5	7	0	0	0	4	2	3
NEB27	+	15	0	4	8	10	1	0	0	0	1	4
NEB28	+	42	8	20	0	11	2	0	3	9	2	5
NEB29	+	19	6	8	0	4	2	0	2	5	2	4
NEB30	+	58	5	39	0	7	3	0	0	9	1	2
NEB31	+	50	8	34	6	7	6	3	0	3	3	8
NEB32	+	54	9	37	0	2	6	3	4	9	3	7
NEB33	+	49	15	27	0	6	8	0	5	8	2	9
NEB34	+	36	5	21	0	5	7	0	1	3	2	3
NEB35	+	26	3	13	4	6	0	0	5	7	3	5
NEB36	+	18	3	9	1	4	2	0	0	3	2	2
NEB37	+	31	8	19	0	1	5	2	4	6	3	7
NEB38	+	24	4	14	2	6	0	0	1	4	3	4
NEB39	+	23	3	11	0	1	6	1	2	5	3	3
NEB40	+	72	12	29	0	8	6	2	14	29	3	14
NEB41	+	16	4	7	2	3	0	0	2	3	3	4
NEB42	+	30	8	25	0	0	5	0	0	0	1	4
NEB43	+	20	2	6	2	5	3	0	0	6	2	2
NEB44	+	24	6	13	1	7	0	0	0	4	2	4
NEB45	+	27	6	21	2	3	0	0	0	3	2	4
NEB46	+	28	3	15	2	7	2	0	0	4	2	2
NEB47	+	26	0	8	0	3	0	0	2	15	1	1
NEB48	+	32	4	12	0	6	2	0	5	12	2	4
NEB49	+	40	6	24	0	1	4	0	2	11	2	4

[1]Scheme in Fig. 1A. NEBs (n=23 from 2 mice after mock injury, n=26 from 3 mice after naphthalene injury) were identified in 20µm cryosections by confocal microscopy.

[2]Either mock (–) or naphthalene (+) airway injury.

[3]The number of distinct Rainbow fluorophores with at least one EdU$^+$ cell present in the NEB (0-4).

[4]Half the total number of EdU$^+$ NE cells present in the NEB regardless of fluorophore identity, rounding down.

Table S2 (related to Figs. 1, S1). Proliferative NE cells in sequentially injured lungs[1]

NEB I.D.	Proliferation marker 1st injury	2nd injury	No. of CGRP+ cells	Prolif. cells 1st injury	Prolif. cells 2nd injury	Prolif. cells Both injuries[2]
NEB50	EdU	BrdU	10	3	2	2
NEB51	EdU	BrdU	40	13	12	9
NEB52	EdU	BrdU	21	9	1	1
NEB53	EdU	BrdU	24	3	6	2
NEB54	EdU	BrdU	104	19	19	16
NEB55	EdU	BrdU	47	12	6	6
NEB56	BrdU	EdU	43	10	1	1
NEB57	BrdU	EdU	26	11	0	0
NEB58	BrdU	EdU	12	1	0	0
NEB59	BrdU	EdU	15	3	0	0
NEB60	BrdU	EdU	28	7	5	5
NEB61	BrdU	EdU	34	11	8	8
NEB62	BrdU	EdU	33	10	14	8
NEB63	EdU	Ki67	11	1	0	0
NEB64	EdU	Ki67	21	2	1	0
NEB65	EdU	Ki67	38	9	0	0
NEB66	EdU	Ki67	28	9	2	2
NEB67	EdU	Ki67	11	0	0	0
NEB68	EdU	Ki67	47	11	3	2
NEB69	EdU	Ki67	76	24	2	2
NEB70	EdU	Ki67	9	2	0	0
NEB71	EdU	Ki67	20	5	2	1
NEB72	EdU	Ki67	19	5	1	1
NEB73	EdU	Ki67	9	5	1	1
NEB74	EdU	Ki67	78	30	6	4
NEB75	EdU	Ki67	28	6	2	0

[1]Schemes in Figs. 1l, S1M. NEBs (n=6 from 2 mice after EdU/BrdU; n=7 from 2 mice after BrdU/EdU; n=13 from 2 mice after EdU/Ki67) were identified in 20µm cryosections by confocal microscopy.

[2]Cells that proliferated after both injuries are also included in the values given for single injuries.

Table S4 (related to Fig. 5). Cell proliferation and Notch activation in NEBs one week after airway injury[1]

NEB I.D.	Injury[2]	No. of CGRP$^+$ cells	Proliferation EdU$^+$	Notch act. Hes1$^+$	Double positive[3]
NEB76	–	35	n.d.[4]	0	n.d.
NEB77	–	2	n.d.	0	n.d.
NEB78	–	25	n.d.	0	n.d.
NEB79	–	50	n.d.	0	n.d.
NEB80	–	45	n.d.	0	n.d.
NEB81	–	17	n.d.	0	n.d.
NEB82	–	14	n.d.	0	n.d.
NEB83	–	26	n.d.	0	n.d.
NEB84	–	2	n.d.	0	n.d.
NEB85	–	2	n.d.	0	n.d.
NEB86	–	38	n.d.	0	n.d.
NEB87	–	18	n.d.	0	n.d.
NEB88	–	10	n.d.	0	n.d.
NEB89	–	9	n.d.	0	n.d.
NEB90	–	29	0	0	0
NEB91	–	40	2	0	0
NEB92	–	31	0	0	0
NEB93	–	18	0	0	0
NEB94	–	1	0	0	0
NEB95	–	1	0	0	0
NEB96	–	80	0	0	0
NEB97	–	13	0	0	0
NEB98	–	42	0	0	0
NEB99	–	15	0	0	0
NEB100	–	15	0	0	0
NEB101	–	32	0	0	0
NEB102	–	29	0	0	0
NEB103	–	39	0	0	0
NEB104	–	5	0	0	0
NEB105	–	28	0	0	0
NEB106	–	21	0	0	0
NEB107	+	37	n.d.	0	n.d.
NEB108	+	33	n.d.	1	n.d.
NEB109	+	36	n.d.	0	n.d.
NEB110	+	8	n.d.	1	n.d.
NEB111	+	32	n.d.	0	n.d.
NEB112	+	2	n.d.	0	n.d.
NEB113	+	37	n.d.	0	n.d.
NEB114	+	10	n.d.	0	n.d.
NEB115	+	8	n.d.	0	n.d.
NEB116	+	5	n.d.	0	n.d.
NEB117	+	14	n.d.	1	n.d.
NEB118	+	11	n.d.	0	n.d.
NEB119	+	15	n.d.	0	n.d.
NEB120	+	3	n.d.	0	n.d.
NEB121	+	22	0	0	0
NEB122	+	35	6	1	0
NEB123	+	12	0	0	0
NEB124	+	24	7	2	0
NEB125	+	5	2	0	0
NEB126	+	20	7	1	1
NEB127	+	21	0	2	0
NEB128	+	4	0	0	0
NEB129	+	34	10	1	0
NEB130	+	6	0	0	0

NEB131	+	9	0	0	0
NEB132	+	16	2	0	0
NEB133	+	20	5	0	0
NEB134	+	7	2	0	0
NEB135	+	15	3	0	0
NEB136	+	12	2	0	0
NEB137	+	10	5	0	0
NEB138	+	36	12	2	0
NEB139	+	25	4	0	0
NEB140	+	21	10	2	2
NEB141	+	16	2	0	0
NEB142	+	24	7	1	0
NEB143	+	6	0	0	0
NEB144	+	22	6	0	0
NEB145	+	8	2	2	2
NEB146	+	19	10	3	2
NEB147	+	24	6	0	0
NEB148	+	23	7	1	1
NEB149	+	2	0	0	0
NEB150	+	11	2	1	0

[1]Scheme in Fig. 5A. NEBs (n=31 from 2 mice after mock injury; n=44 from 2 mice after naphthalene injury) were identified in 20µm cryosections by confocal microscopy.

[2]Either mock (−) or naphthalene (+) airway injury.

[3]Double-positive cells are also included in single-positive values.

[4]n.d., not determined

Table S5 (related to Fig. 6). Proliferative NE cells in sequentially injured and *Rb/p53*-deleted lungs[1]

NEB I.D.	Proliferation marker Injury	ΔRb/p53	No. of $CGRP^+$ cells	Prolif. cells Injury	Prolif. cells ΔRb/p53	Prolif. cells Injury & ΔRb/p53[2]
NEB151	EdU	BrdU	27	5	3	2
NEB152	EdU	BrdU	45	0	0	0
NEB153	EdU	BrdU	18	3	7	3
NEB154	EdU	BrdU	28	7	7	5
NEB155	EdU	BrdU	29	9	8	6
NEB156	EdU	BrdU	26	9	4	4
NEB157	BrdU	EdU	28	6	3	3
NEB158	BrdU	EdU	24	4	1	0
NEB159	BrdU	EdU	20	3	3	2
NEB160	BrdU	EdU	12	3	0	0
NEB161	BrdU	EdU	13	2	1	1
NEB162	BrdU	EdU	19	0	0	0
NEB163	BrdU	EdU	20	2	0	0
NEB164	BrdU	EdU	24	3	1	1
NEB165	BrdU	EdU	34	15	3	2
NEB166	BrdU	EdU	29	8	0	0
NEB167	BrdU	EdU	18	3	3	3
NEB168	BrdU	EdU	69	10	6	2

[1]Scheme in Fig. 6l. NEBs (n=6 from 1 mouse after EdU/BrdU; n=12 from 2 mice after BrdU/EdU) were identified in 20µm cryosections by confocal microscopy.

[2]Cells that proliferated under both conditions are also included in values given for single conditions.